











NCBI RefSeq Resources for Plant Genomics

Anjana R Vatsan(raina@nih.gov)

Functional Genomics Workshop

PAG XXVIII January 13, 2020



Also from NCBI!

Visit NCBI Booth 321 Contact us info@ncbi.nlm.nih.gov

Day	Time	Topic
Monday	12:50 pm – 3:00 pm <i>Pacific Salon 1</i>	NCBI Genome Resources Workshop
Tuesday	11:10 am California	NCBI BLAST: Enhanced Web Usability through New Result Page and Effective Genomic Data Access Digital Tools and Resources Session 3
Wednesday	11:50 am <i>California</i>	Federated Cloud Access to Datasets through Indexing and/or Graphs! Digital Tools and Resources Session 4





NCBI Genome Resources Workshop

Monday January 13, 2020, 12:50 – 3:00 pm, Pacific Salon 1

Time	Topic
12:55 – 1:15	NCBI Wants Your Sequence Data! How Do I Get It There? Ilene Mizrachi
1:15 – 1:35	Annotation of Eukaryote Genomes at NCBI Jinna Hoffman
1:35 – 1:55	Accessing Homologous Gene Datasets at NCBI Nuala O'Leary
1:55 – 2:15	The New PubMed Is Here! Kathi Canese
2:15 – 2:35	Taxonomy Lookup; Data Retrieval: How to Find and Stream Genomic Data in the Cloud! Ben Busby

Visit NCBI Booth 321

Contact us info@ncbi.nlm.nih.gov









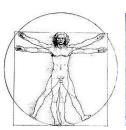






















NCBI RefSeq Resources for Plant Genomics

Anjana R Vatsan(raina@nih.gov)

Functional Genomics Workshop

PAG XXVIII January 13, 2020



RefSeq: NCBI Reference Sequence Database

RefSeq is a public database of nucleotide and protein sequences which are derived, in most part, from genome assemblies that are submitted to International Nucleotide Sequence Database Collaboration (INSDC), by one of the following methods.

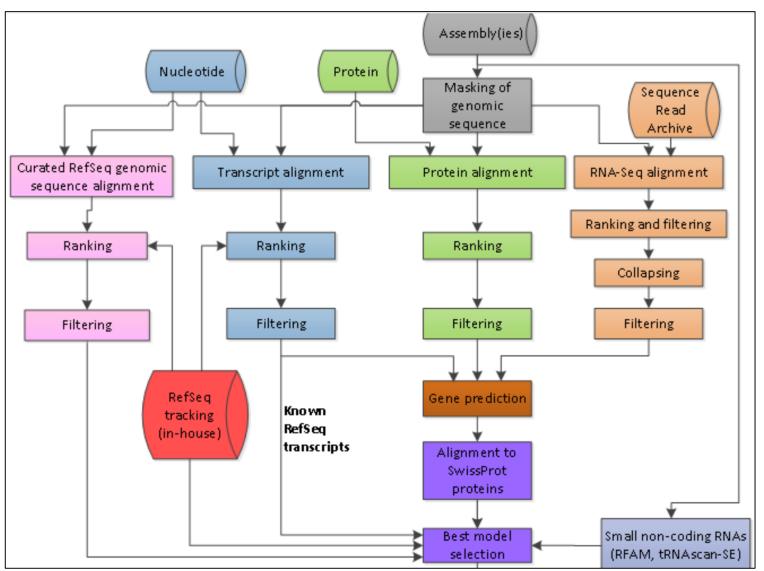
- Computationally using the Genome Annotation Pipeline
- Manual curation
- Propagation from model organism databases
 e.g. Arabidopsis thaliana

https://www.ncbi.nlm.nih.gov/refseq/





Annotation Pipeline

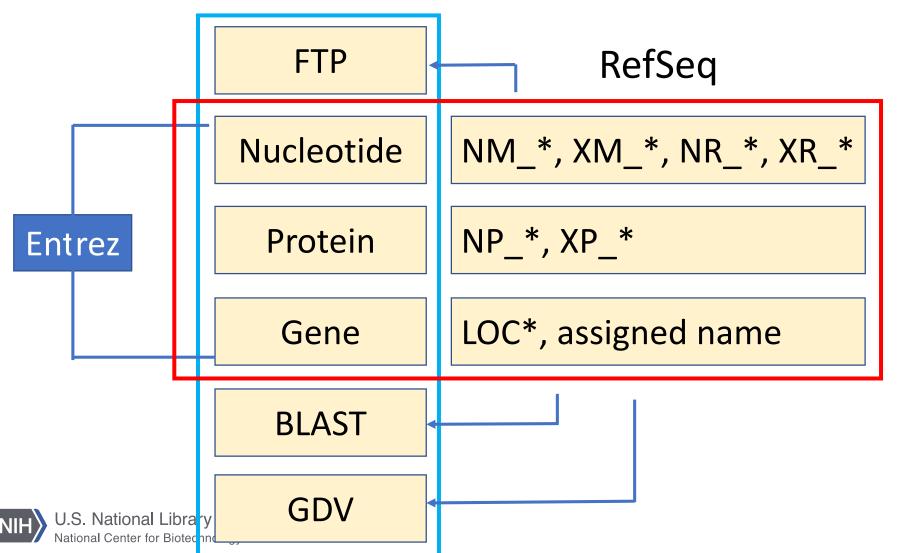






Annotation Output

ftp://ftp.ncbi.nlm.nih.gov/genomes/refseq/plant/





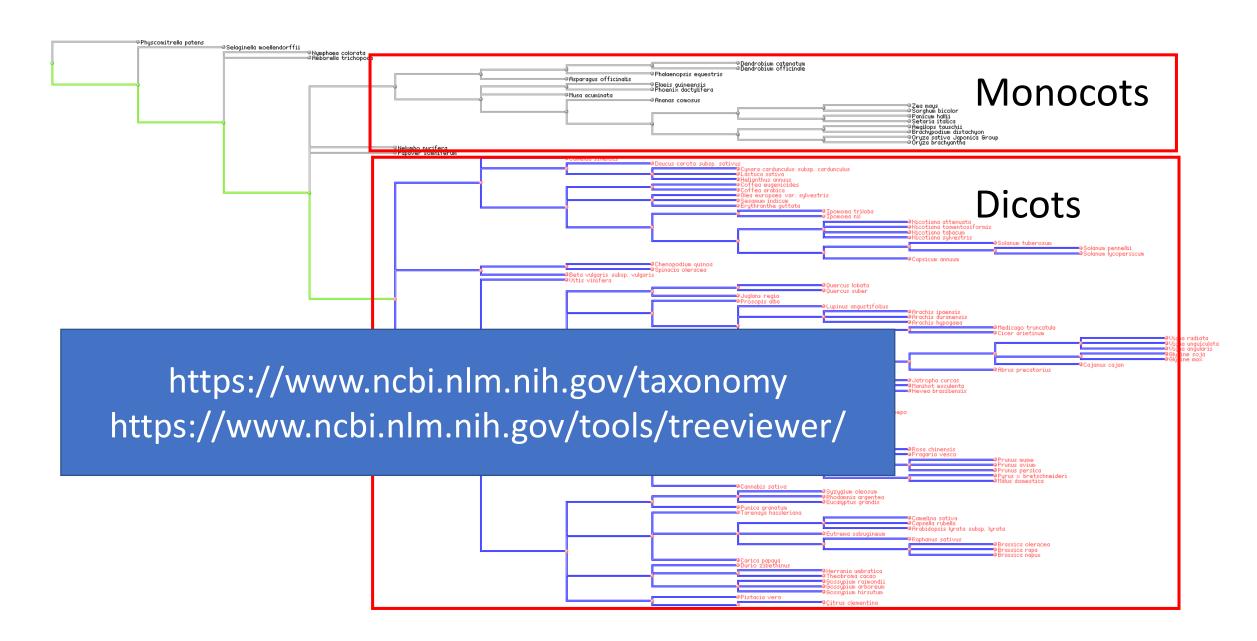
RefSeq Data organization



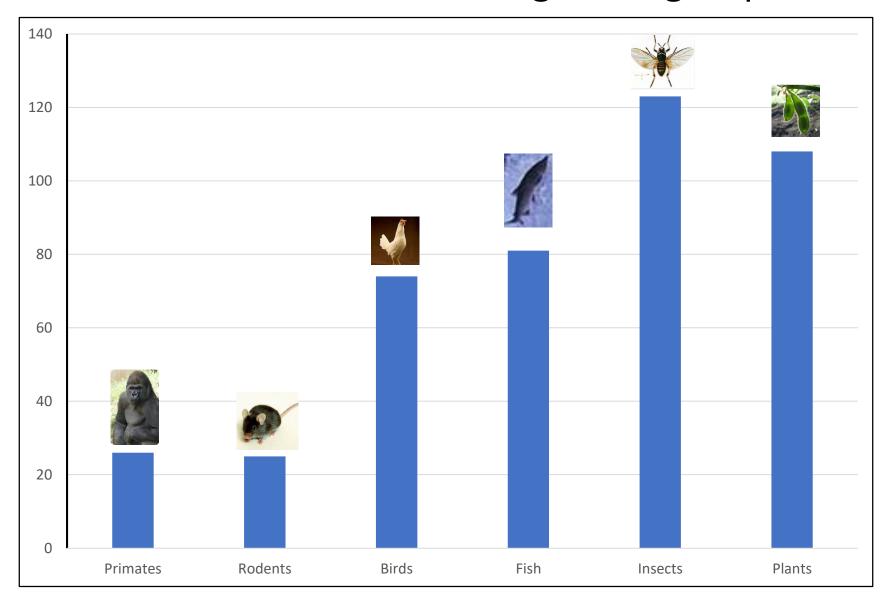




108 plant genomes have been annotated at NCBI



Annotation of various organism groups



Assembly

soybean[orgn]

Best representative assembly chosen for annotation

Glycine max vz.1

1. Organism: Glycine max (soybean)

Infraspecific name: Cultivar: Williams 82

Submitter: US DOE Joint Genome Institute (JGI-PGF)

Date: 2018/07/24

Assembly level: Chromosome Genome representation: full

RefSeq category: representative genome

GenBank assembly accession: GCA_000004515.4 (latest) RefSeq assembly accession: GCF_000004515.5 (latest) IDs: 1832791 [UID] 6943708 [GenBank] 7001488 [RefSeq]

Glycine max Enrei 2.0

 Organism: Glycine max (soybean) Infraspecific name: Cultivar: ENREI

Submitter: National Institute of Agrobiological Sciences

Date: 2015/08/06 Assembly level: Contig Genome representation: full

GenBank assembly accession: GCA 001269945.2 (latest)

RefSeq assembly accession: n/a

IDs: 474001 [UID] 2242528 [GenBank]

□ glyma.Lee.gnm1

Organism: **Glycine max** (soybean)
Infraspecific name: Cultivar: Lee

Submitter: Glycine max cv Lee and Glycine soja PI 483463 sequencing consortium

Access assembly meta-data,

genomic sequence data

statistical reports, and links to

Date: 2018/01/30

Assembly level: Chromosome Genome representation: full

GenBank assembly accession: GCA_002905335.2 (latest)

RefSeq assembly accession: n/a IDs: 2580391 [UID] 9338298 [GenBank]

☐ Gmax ZH13

Organism: Glycine max (soybean)

Infraspecific name: Cultivar: Zhonghuang 13

Submitter: Institute of Genetics and Developmental Biology , Chinese Academy of Science

Date: 2018/08/10





Annotation Home Page

Eutrema salsugineum (saltwater cress)	<u>Eutsalg1_0</u> (GCF_000478725.1)	100	2018-02-22	2018-02-26	FTP	В	AR	GDV
Fragaria vesca (wild strawberry)	FraVesHawaii_1.0 (GCF_000184155.1)	101	2015-03-02	2015-03-04	FTP	В	AR	GDV
Glycine max (soybean)	Glycine_max_v2.1 (GCF_000004515.5)	103	2018-08-07	2018-09-06	FTP	В	AR	GDV
Glycine soja (wild soybean)	ASM419377v2 (GCF_004193775.1)	100	2019-03-08	2019-03-12	FTP	В	AR	GDV
Gossypium arboreum (tree cotton)	Gossypium_arboreum_v1.0 (GCF_000612285.1)	100	2016-08-01	2016-08-11	FTP	В	AR	GDV
Gossypium hirsutum (cotton)	ASM98774v1 (GCF_000987745.1)	100	2016-05-09	2016-05-18	FTP	В	AR	GDV
Gossypium raimondii (eudicots)	<u>Graimondii2_0</u> (GCF_000327365.1)	100	2015-04-29	2015-05-22	FTP	В	AR	GDV
Helianthus annuus (common sunflower)	HanXRQr1.0 (GCF_002127325.1)	100	2017-07-28	2017-08-07	FTP	В	AR	GDV
Herrania umbratica (eudicots)	ASM216827v2 (GCF_002168275.1)	100	2017-06-08	2017-06-09	FTP	В	AR	GDV
Hevea brasiliensis (rubber tree)	ASM165405v1 (GCF_001654055.1)	100	2017-07-16	2017-07-19	FTP	В	AR	GDV
Ipomoea nil (Japanese morning glory)	<u>Asagao_1.1</u> (GCF_001879475.1)	100	2016-11-25	2016-11-29	FTP	В	AR	GDV
Ipomoea triloba (trilobed morning glory)	ASM357664v1 (GCF_003576645.1)	100	2019-10-11	2019-10-17	FTP	В	AR	GDV
Jatropha curcas (eudicots)	<u>JatCur_1.0</u> (GCF_000696525.1)	101	2017-03-31	2017-04-06	FTP	В	AR	GDV





Feature	Glycine_max_v2.1			
Genes and pseudogenes ⁽²⁾	59,906			
protein-coding	46,993			
non-coding	7,881			
transcribed pseudogenes	376			
non-transcribed pseudogenes	4,656			
genes with variants	13,958			
immunoglobulin/T-cell receptor gene segments	0			
other	0			
mRNAs	71,048			
fully-supported	64,943			
with > 5% ab initio ⊌	5,225			
partial	410			
with filled gap(s) ⁽²⁾	37			
known RefSeq (NM_) ⁽²⁾	7,593			
model RefSeq (XM_)	63,455			
non-coding RNAs [®]	14,358			
fully-supported	11,288			
with > 5% ab initio ¹	0			

Annotation Report

Detailed reports

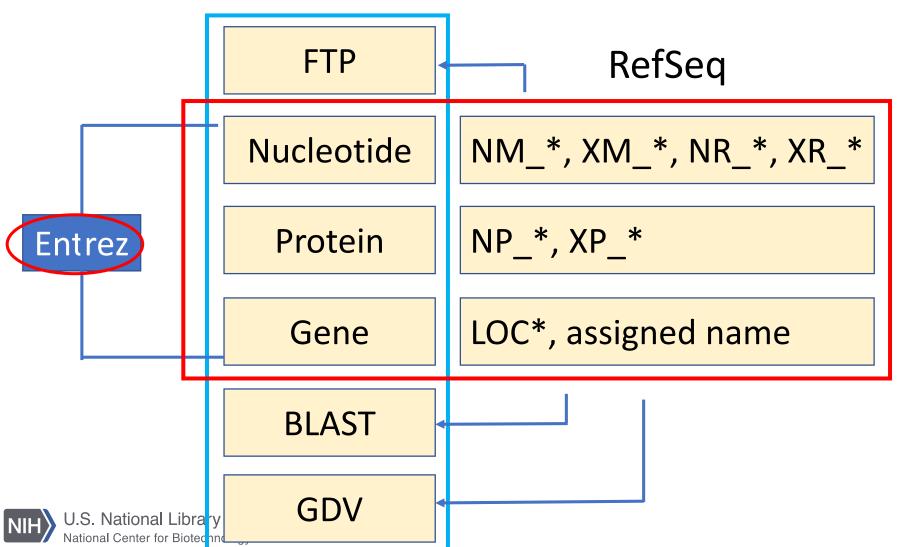
Feature	Count	Mean length (bp)	Median length (bp)	Min length (bp)	Max length (bp)
Genes	54,874	4,017	2,958	54	287,040
All transcripts	85,406	1,872	1,655	18	16,833
mRNA	71,048	1,953	1,712	148	16,833
misc_RNA	4,407	2,389	2,080	197	8,834
miRNA	614	22	21	18	25
tRNA	752	74	73	71	93
IncRNA	6,360	1,521	1,146	54	11,067
snoRNA	1,687	105	107	64	228
snRNA	136	151	158	100	198
rRNA	402	1,615	1,807	104	3,470
Single-exon transcripts	7,813	1,274	1,044	148	8,719
coding transcripts (NM_/XM_)	7,780	1,275	1,046	148	8,719

https://www.ncbi.nlm.nih.gov/genome/annotation_euk/all

partial 0

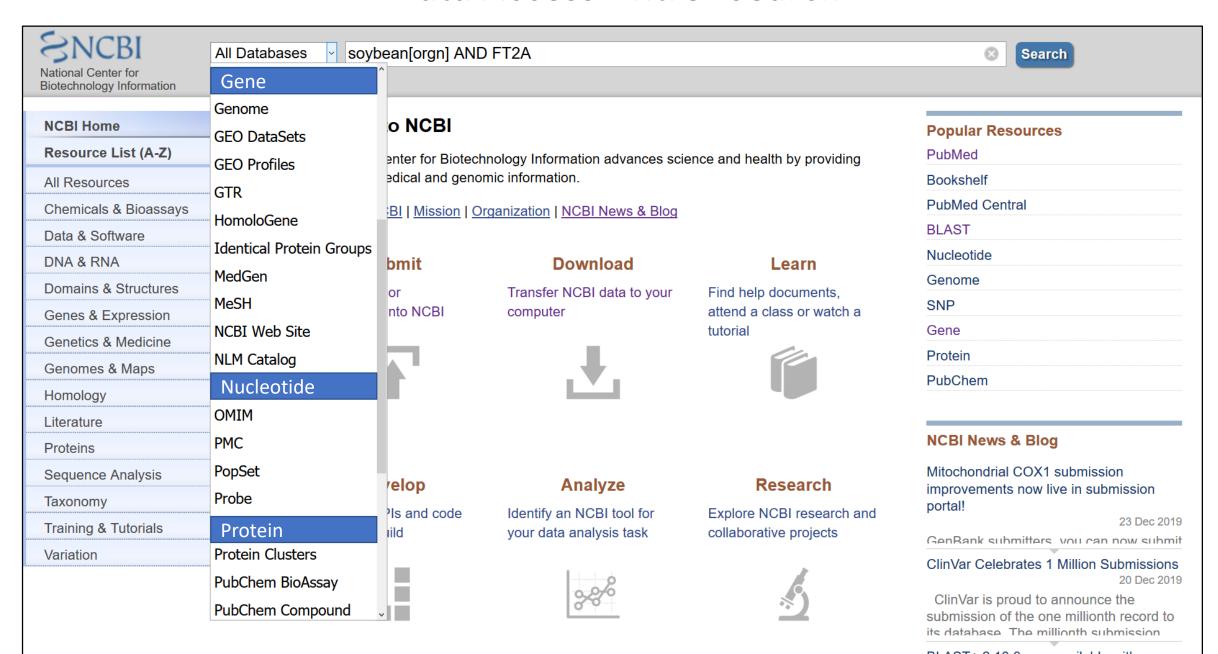
Annotation Output

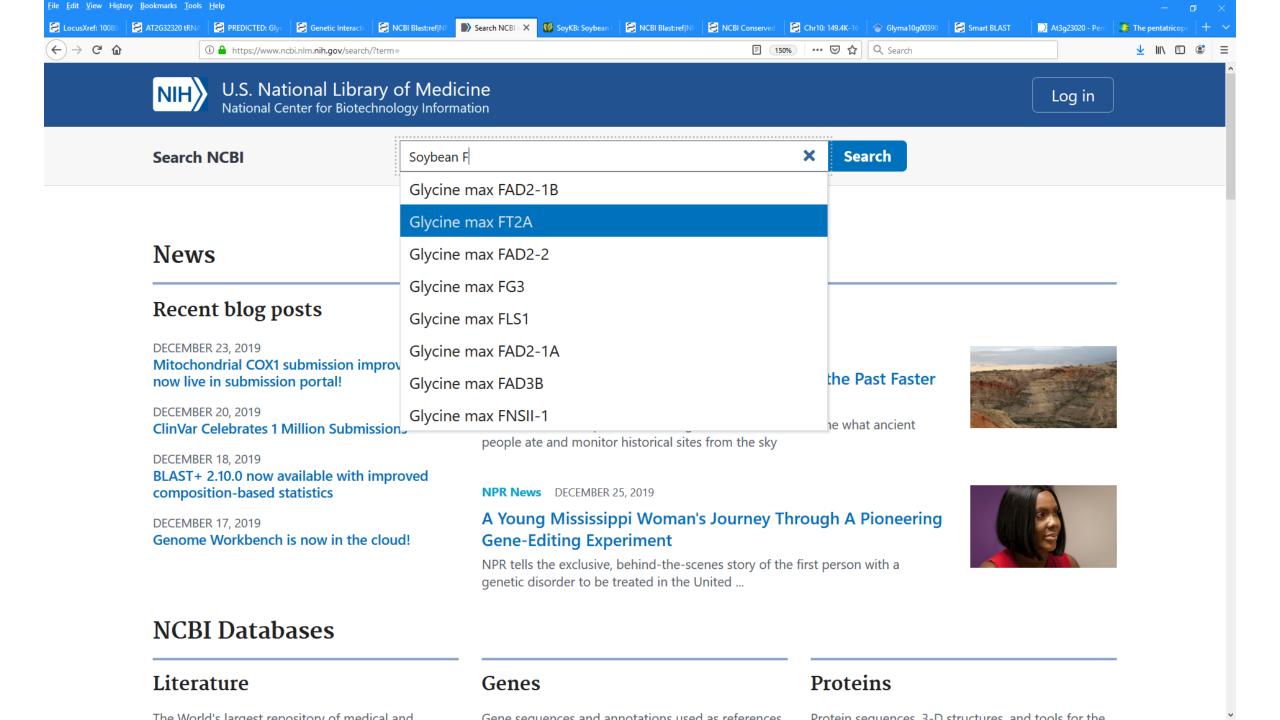
ftp://ftp.ncbi.nlm.nih.gov/genomes/refseq/plant/

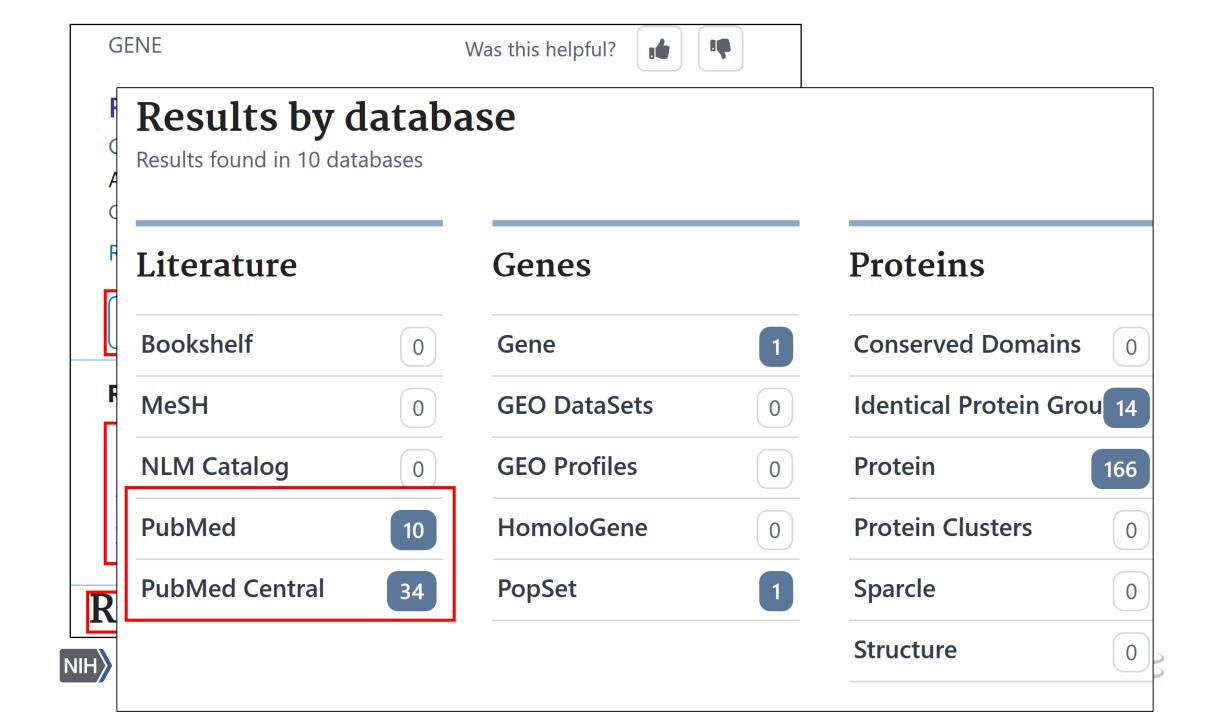




Data Access-Entrez search











FT2A – protein FLOWERING LOCUS T

Glycine max (soybean)

Also known as: GLYMA_16G150700, E9, FT, FT3, FTL3, GmFT2a

GenelD: 100814951

RefSeq transcripts (2) RefSeq proteins (2) PubMed (10)

Genome Browser

BLAST

Download

RefSeq Sequences

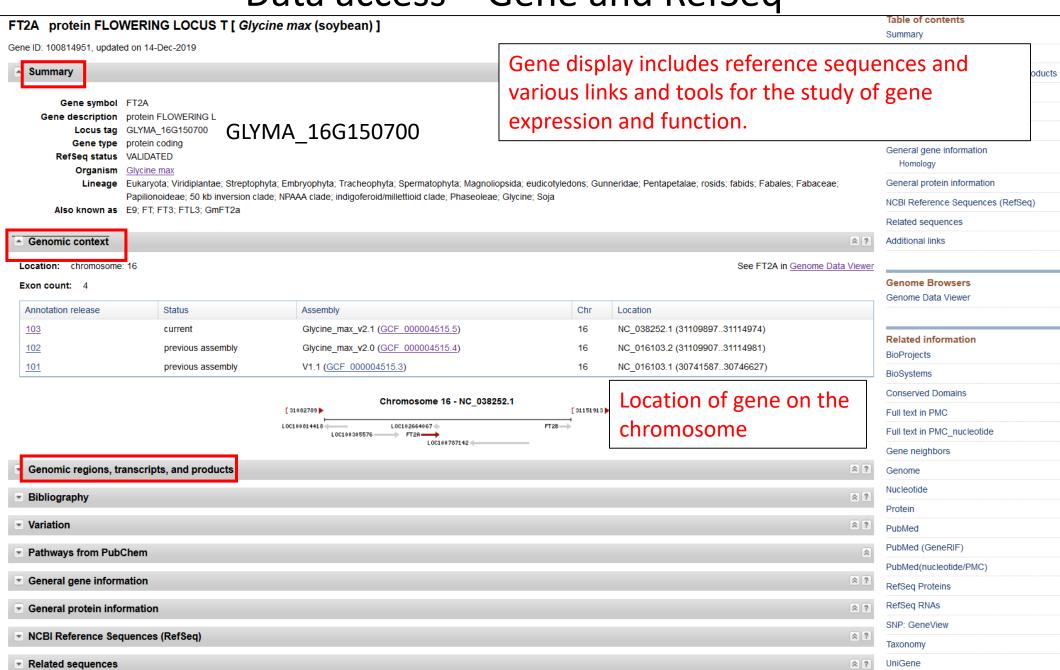
NM_001253256.2 899 NP_001240185.1 176

XM_006598696.3 920 XP_006598759.1 158 X1

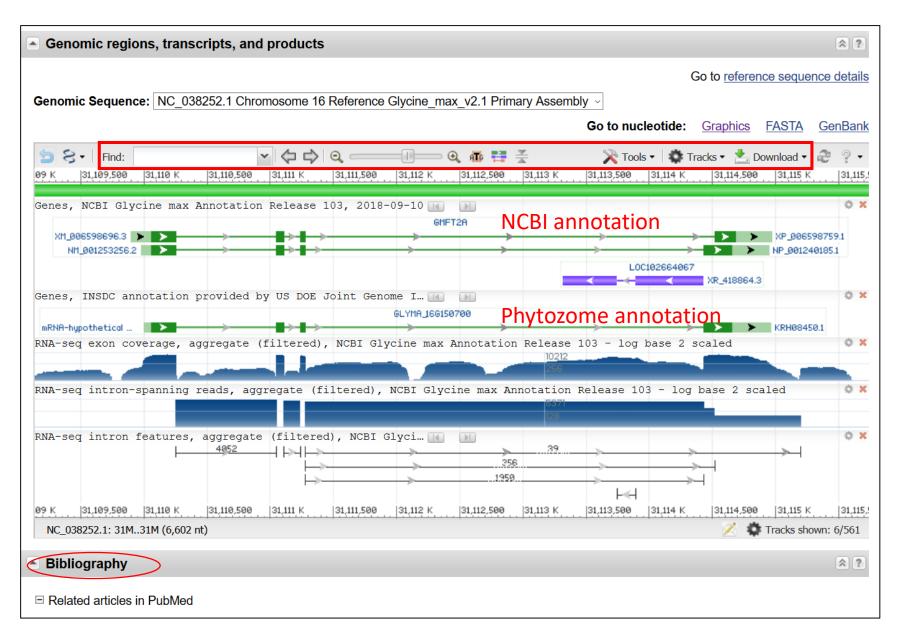
Results by database



Data access – Gene and RefSeq











Bibliography

□ Related articles in PubMed

- 1. CRISPR/Cas9-mediated targeted mut Cai Y, et al. Plant Biotechnol J, 2018 Jan.
- 2. GmFT2a polymorphism and maturity Jiang B, et al. PLoS One, 2013. PMID 241
- 3. GmFT2a, a soybean homolog of FLOV Sun H, et al. PLoS One, 2011. PMID 2219
- 4. Mutagenesis of GmFT2a and GmFT5a Cai Y, et al. Plant Biotechnol J, 2020 Jan.
- 5. Functional divergence between soybe Takeshima R, et al. J Exp Bot, 2019 Aug 7

See all (10) citations in PubMed

☐ GeneRIFs: Gene References Into Function

What's a GeneRIF?

- 1. GmFT1a expression was induced by I expression of flowering promoters Gm
- 2. Expression of the FLOWERING LOCU
- 3. Although GmFT2a is a key flowering g
- 4. GmFT2a expression is associated with

Submit: New GeneRIF

Correction

GeneRIE (Con Unto Eupotion) anables interacted ecientists to enrich the functional

Feedback for Gene and Reference Sequences (RefSeq) **NCBI**

Make suggestions, submit additions and corrections, or ask for help concerning Gene or Reference Sequence (RefSeg) records. See additional information: Gene Home Page, RefSeg Home Page.

Do not use this form to report a problem in PubMed or GenBank. Do not use this form to submit sequence data to NCBI.

Additional contacts:

annota

and th annota

Please

will not

For ad

To sug please

Each (

GeneF

Submi

Gene

10081

PubMe

GeneF

- PubMed report typographical or other errors in citations
- GenBank submission documentation submit sequence data to GenBank
- NCBI help contact us about other NCBI resources

What would you like to do?

Return to previous page

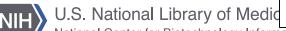
Add a GeneRIF - Add a publication with a functional comment to a Gene record.

Additions

- Report a new gene that is not yet available in Gene
- Request addition of a RefSeg transcript, protein, or pseudogene record
- Contribute a summary describing the function of the gene

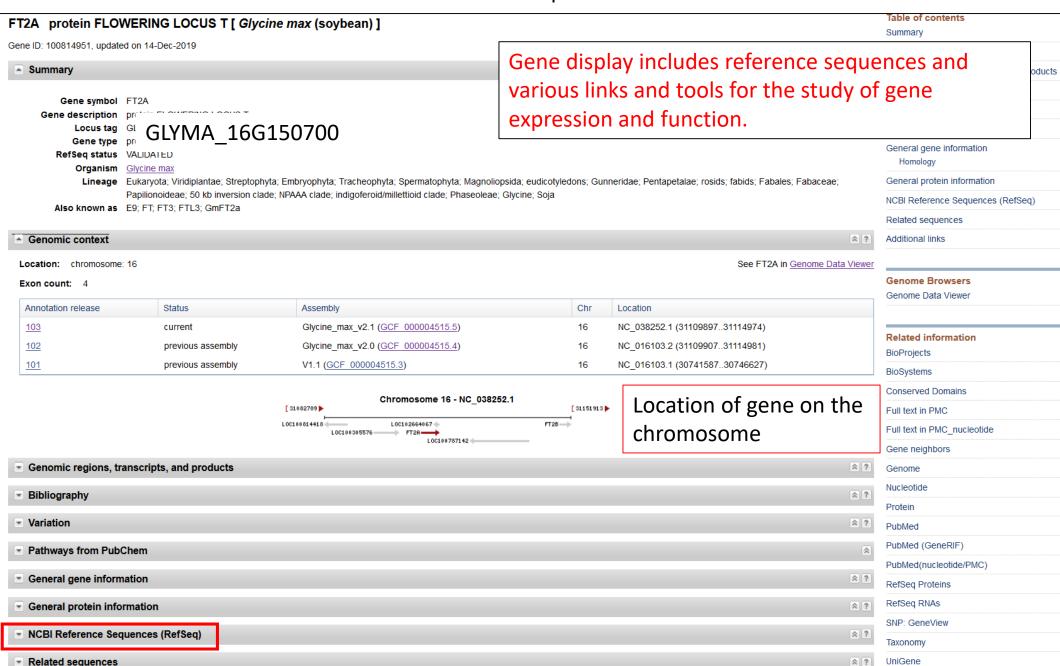
Corrections

- Correct or update a Gene record (please provide the GeneID)
- Correct or update a RefSeg record (please provide the accession.version)
- Report a publication that is incorrectly associated with a Gene or RefSeg (please provide the PubMed ID)



National Center for Biotechnology Information

Data access – Gene and RefSeq





► NCBI Reference Sequences (RefSeq)

☐ RefSeqs maintained independently of Annotated Genomes

These reference sequences exist independently of genome builds. Explain

Status: VALIDATED		
Source sequence(s)	ACUP03010190, KJ607992	
UniProtKB/TrEMBL	E3NYP3	
Conserved Domains (1) si	ummary	
	cl00227 Location:1 → 175	PEBP; PhosphatidylEthanolamine-Binding Protein (PEBP) do
	1	
	0005007504	dylethanolamine-binding protein FT2a isoform X1

Related sequences From GenBank

FT2A has two transcript variants

NM_001253256.2 XM_006598696.3

Difference between N* and X* accessions

- N* accessions are used for known RefSeqs. This category is supported by manual curation. Records are primarily derived from INSDC cDNAs, EST, and Transcript Shotgun Assembly (TSA) records.
- X* accessions are predicted models. This category is computationally predicted based on aligned evidence. Records are primarily derived from genomic sequence. The vast majority of XMs are fully supported by experimental evidence, and for most species they are on par, quality-wise, with the NMs.





NM 001253256.2

XM_006598696.3

```
VALIDATED REFSEQ This record has undergone validation or
COMMENT
            preliminary review. The reference sequence was derived from
            ACUP03010190.1 and KJ607992.1.
            On Jul 19, 2017 this sequence version replaced NM 001253256.1.
            ##Evidence-Data-START##
           Transcript exon combination :: EU287455.1, AB550122.1 [EC0:000033]
           RNAseq introns
                                        :: single sample supports all introns
                                           SAMN02009287, SAMN02215336
                                           [ECO:0000348]
            ##Evidence-Data-END##
PRIMARY
           REFSEQ SPAN
                                PRIMARY IDENTIFIER PRIMARY SPAN
                                                                        COMP
           1 - 77
                                ACUP03010190.1
                                                    61121-61197
            78-608
                                KJ607992.1
                                                    1 - 531
            609-899
                                                    65802-66092
                                ACUP03010190.1
                     Location/Oualifiers
FEATURES
                     1..899
    source
                     /organism="Glycine max"
                     /mol type="mRNA"
                     /cultivar="Williams 82"
                     /db xref="taxon:3847"
                     /chromosome="16"
```

```
COMMENT
           MODEL REFSEQ: This record is predicted by automated computational
           analysis. This record is derived from a genomic sequence
            (NC 038252.1) annotated using gene prediction method: Gnomon.
                Documentation of NCBI's Annotation Process
           On Aug 15, 2018 this sequence version replaced XM 006598696.2.
            ##Genome-Annotation-Data-START##
            Annotation Provider
                                        :: NCBI
                                        :: Full annotation
            Annotation Status
            Annotation Name
                                        :: Glycine max Annotation Release 103
                                        :: 103
            Annotation Version
            Annotation Pipeline
                                        :: NCBI eukaryotic genome annotation
                                           pipeline
            Annotation Software Version :: 8.1
            Annotation Method
                                        :: Best-placed RefSeq; Gnomon
            Features Annotated
                                        :: Gene; mRNA; CDS; ncRNA
            ##Genome-Annotation-Data-END##
FEATURES
                     Location/Oualifiers
                     1..920
    source
                     /organism="Glycine max"
                     /mol type="mRNA"
                    /cultivar="Williams 82"
                    /db xref="taxon:3847"
                     /chromosome="16"
                    /tissue type="callus"
```





Data curation—how do we maintain the quality of our data

PRR3A gene (GeneID: **100785796**)

Low Quality protein, corrected it based on

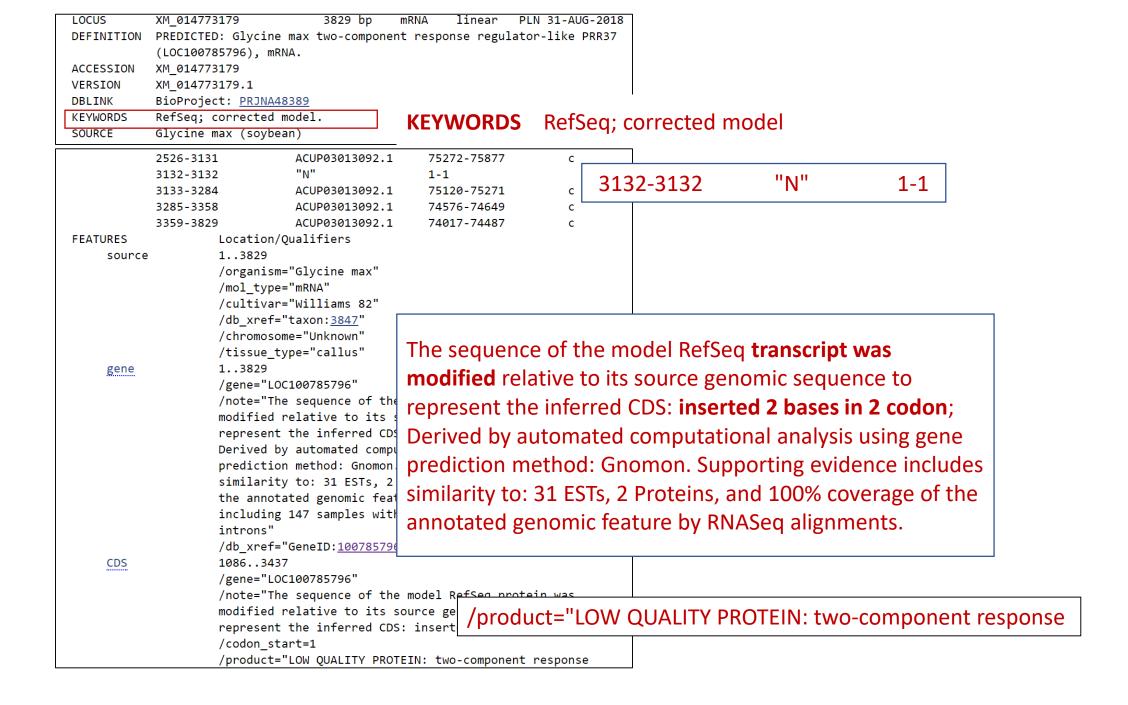
PMID: 30418611*



^{*}Li et. al. Plant Cell Physiol. 2019 Feb 1;60(2):407-420. Characterization of Two Growth Period QTLs Reveals Modification of PRR3 Genes During Soybean Domestication.

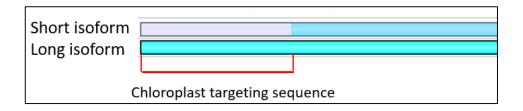




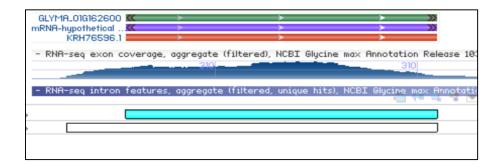


Data curation: Add value based on publications

Adding value to data: Updated HPPD gene (GeneID:100101901) to create two isoforms, short and long, based on PMID: 25192697*.



Updating locus_type: Updated LUXa gene from coding to non-coding based on PMID:28878247**



*Seihl et. al. Plant Physiol. 2014 Nov;166(3):1162-1176. Broad 4-hydroxyphenylpyruvate dioxygenase inhibitor herbicide tolerance in soybean with an optimized enzyme and expression cassette **Liew et. Al. Sci Rep. 2017 Sep 6;7(1):10605. A novel role of the soybean clock gene LUX ARRHYTHMO in male reproductive development.

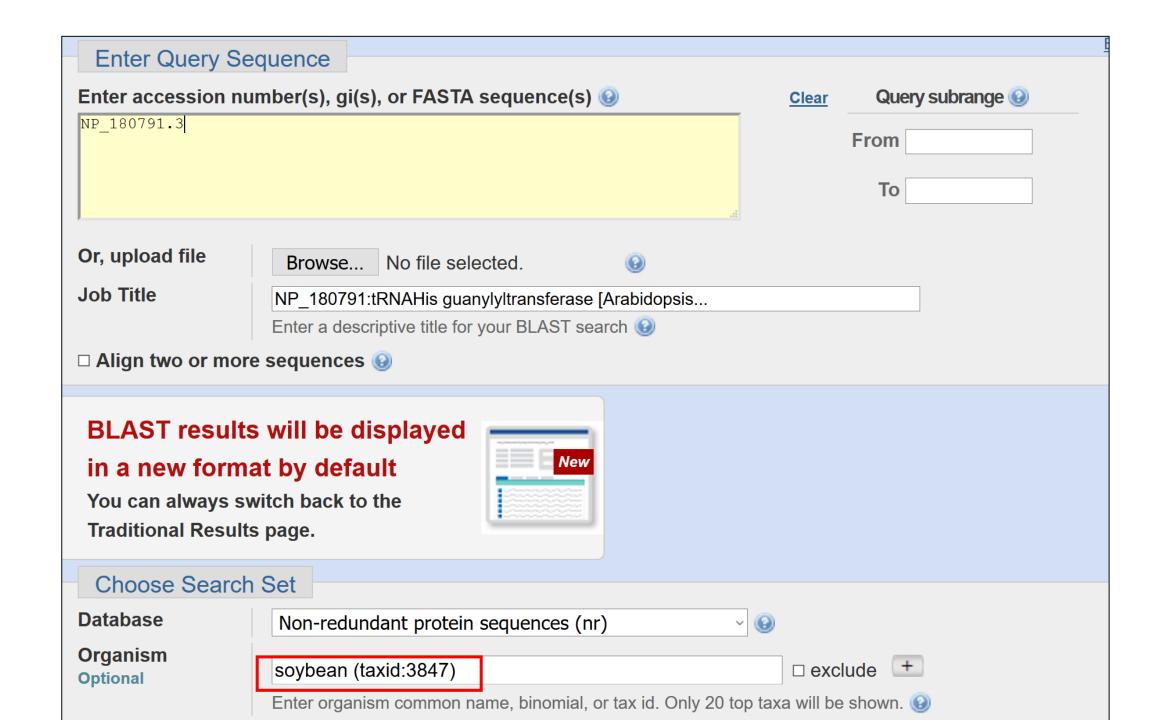




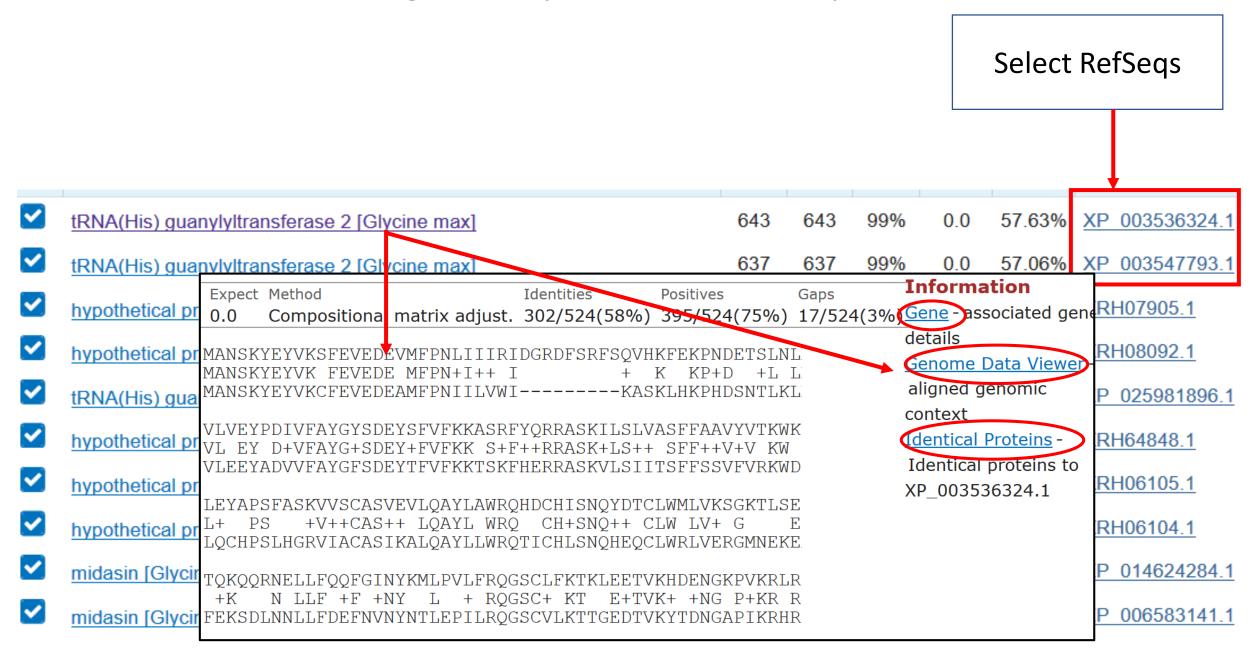
Data Analysis using BLAST

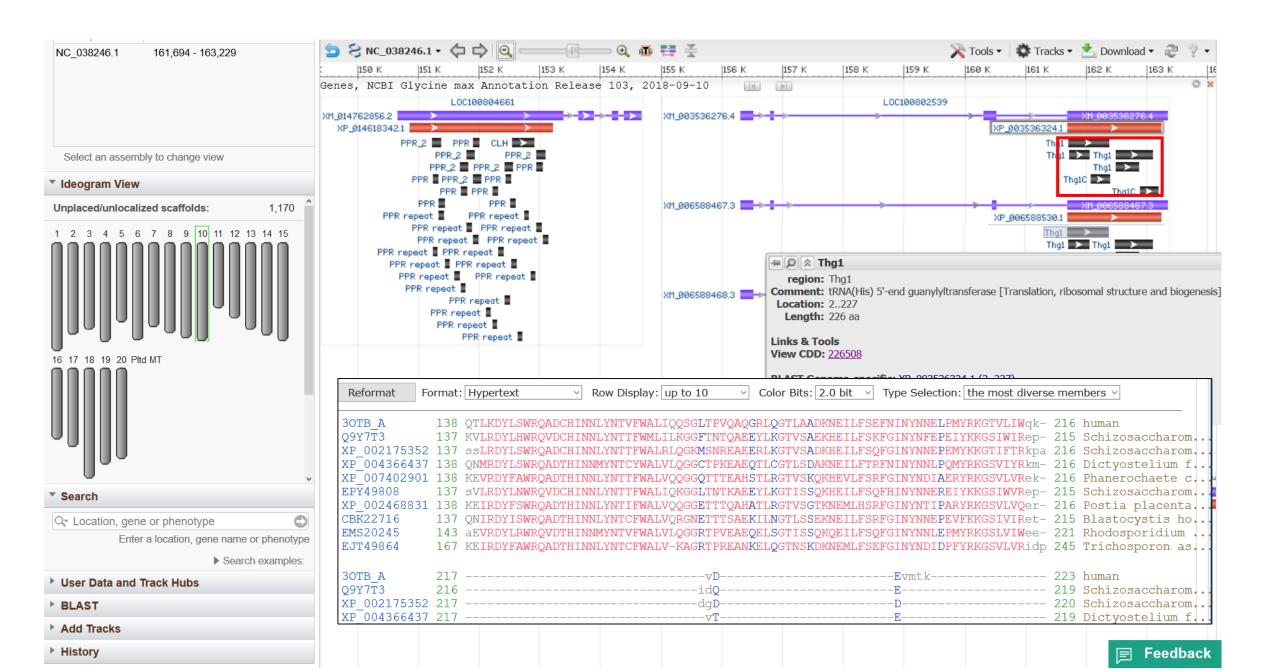
ICA2 gene (AT2G32320) involved in growth and flowering time plasticity in relation to temperature in 1. NM 128791.4 - NP 180791.3 tRNAHis guanylyltransferase [Arabidopsis thaliana] Arabidopsis; PMID: 30992321* See identical proteins and their annotated locations for NP 180791.3 Status: REVIEWED Gene Gene At2g32 UniProtKB/Swiss-Prot | F4ISV6 Create Conserved Domains (2) summary AT2G32320 tRNAHis guanylyltransfer Thg1; tRNAHis guanylyltransferase pfam04446 Location:275 → 401 Gene ID: 817793, updated on 25-Oct-2019 Thg1C; Thg1 C terminal domain pfam14413 Location:405 → 510 Summary 2. NM 001161072.1 → NP 001154544.1 tRNAHis guanylyltransferase [Arabidopsis thaliana] Genomic context See identical proteins and their annotated locations for NP 001154544.1 Genomic regions, transcripts, and products Status: REVIEWED UniProtKB/Swiss-Prot F4ISV6 **Bibliography** Conserved Domains (3) summary Thg1; tRNA(His) 5'-end guanylyltransferase [Translation, ribosomal COG4021 Variation Location:286 → 531 structure and biogenesis] pfam04446 Thg1; tRNAHis guanylyltransferase Pathways from PubChem Location:287 → 413 Thg1C; Thg1 C terminal domain pfam14413 Location: $417 \rightarrow 522$ General gene information 3. NM 001161073.1 → NP 001154545.1 tRNAHis guanylyltransferase [Arabidopsis thaliana] General protein information NCBI Reference Sequences (RefSeq) ☆ ?

^{*}Mendez-Vigo et. al. Plant Cell. 2019 Jun;31(6):1222-1237. Genetic Interactions and Molecular Evolution of the Duplicated Genes *ICARUS2* and *ICARUS1* Help Arabidopsis Plants Adapt to Different Ambient Temperatures.



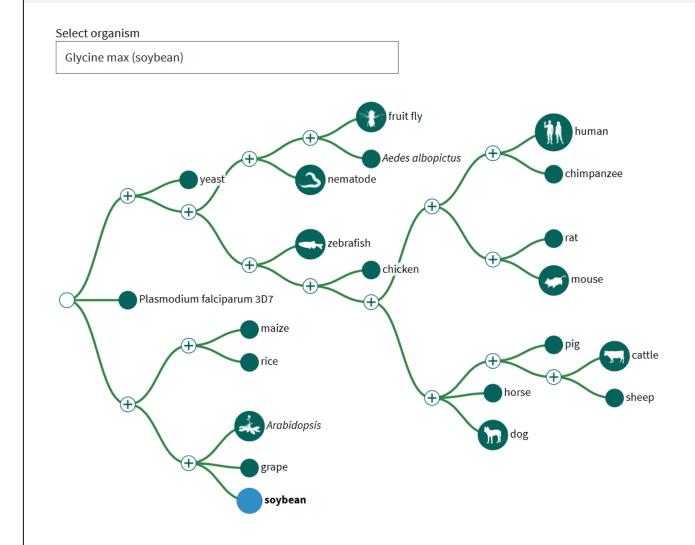
Accessing RefSeq from BLAST output

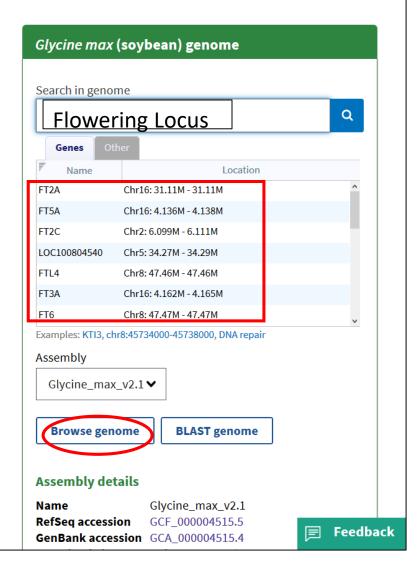


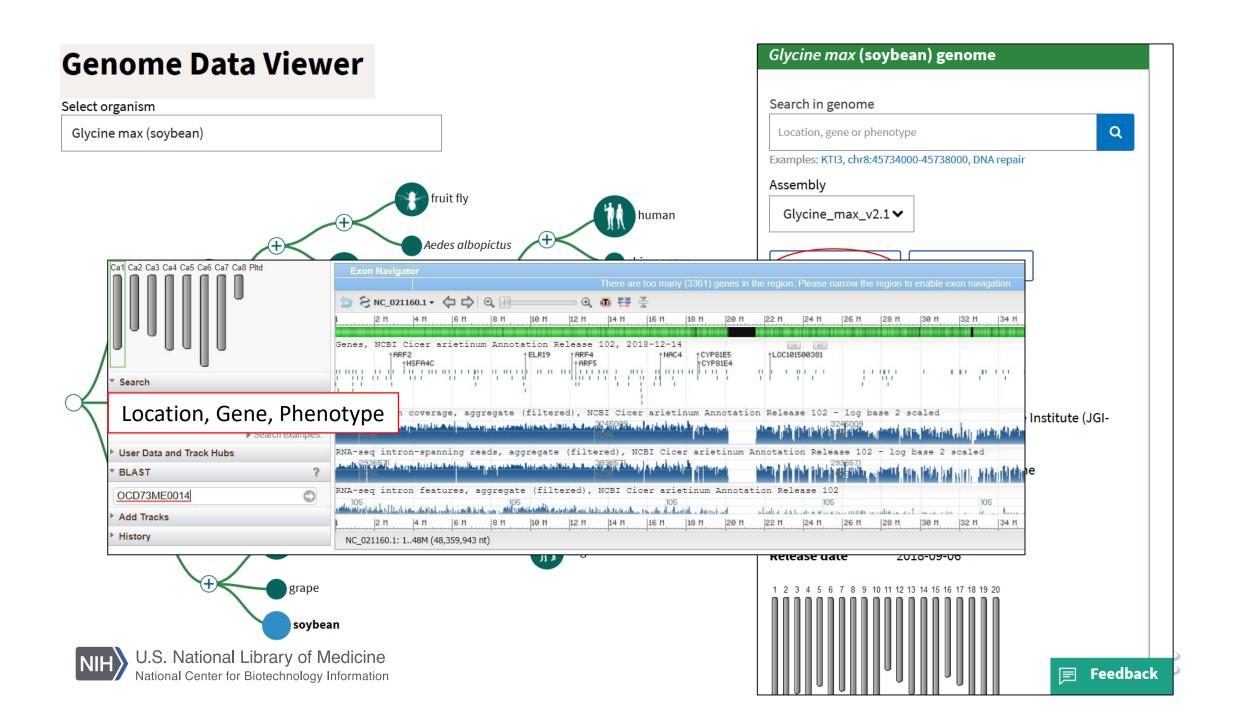


Genome Data Viewer

GDV is a genome browser supporting the exploration and analysis of more than 740 eukaryotic RefSeq genome assemblies. •



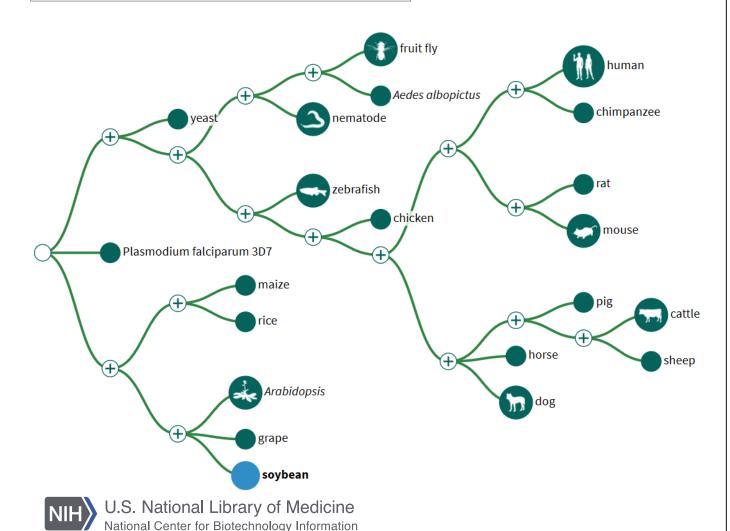


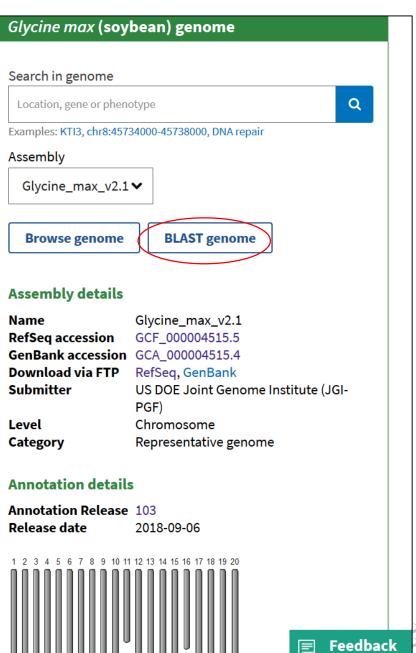


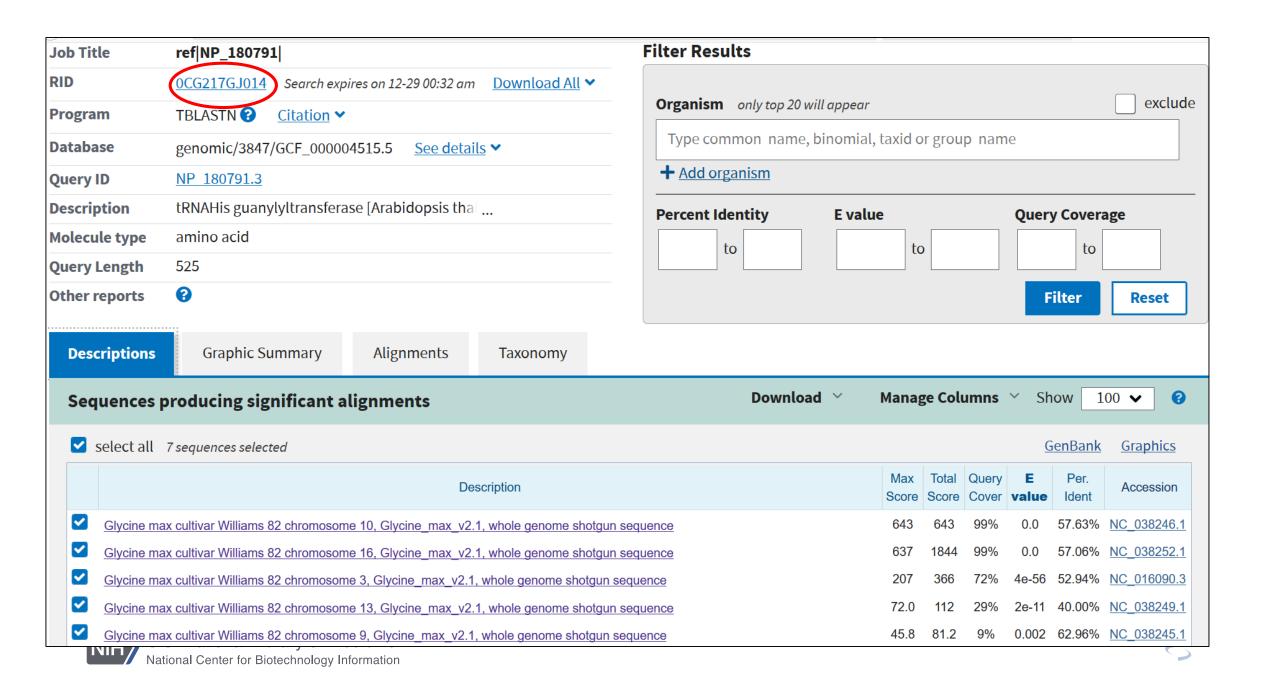
Genome Data Viewer

Select organism

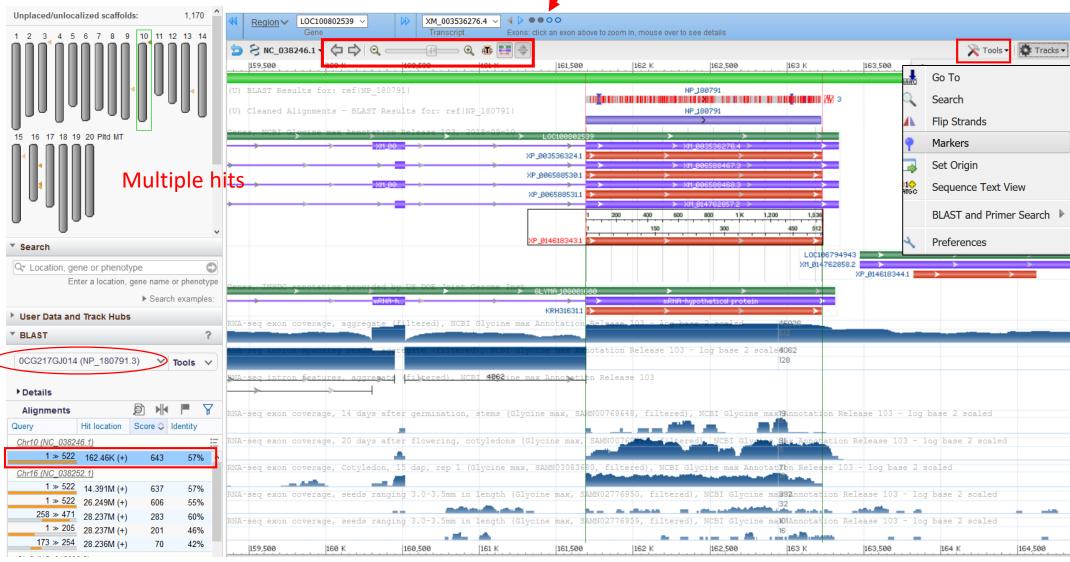
Glycine max (soybean)

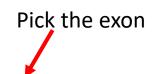


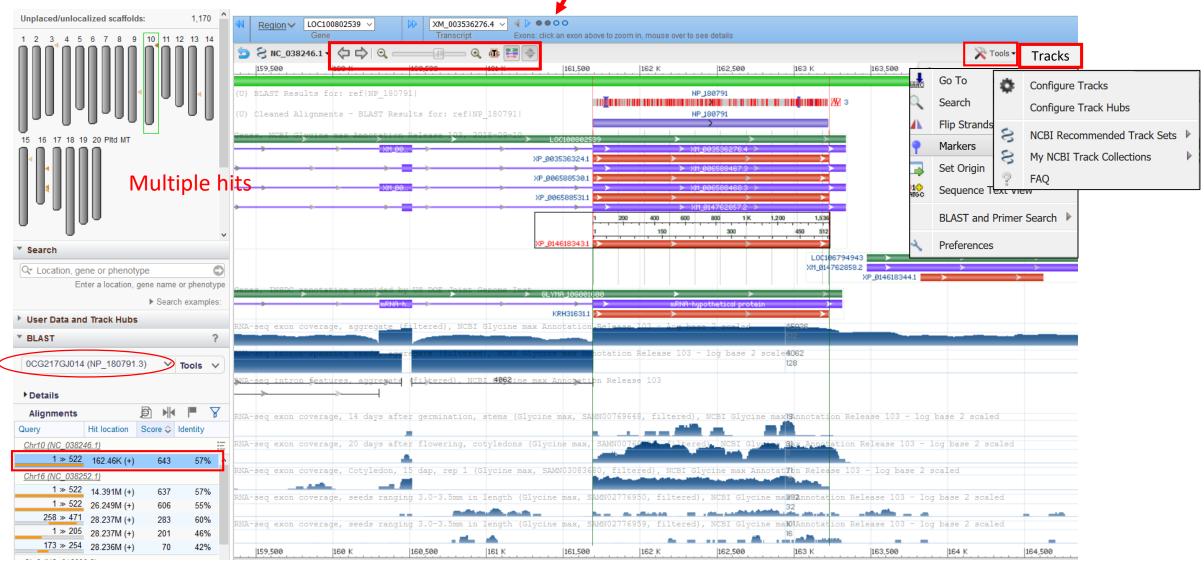


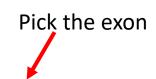


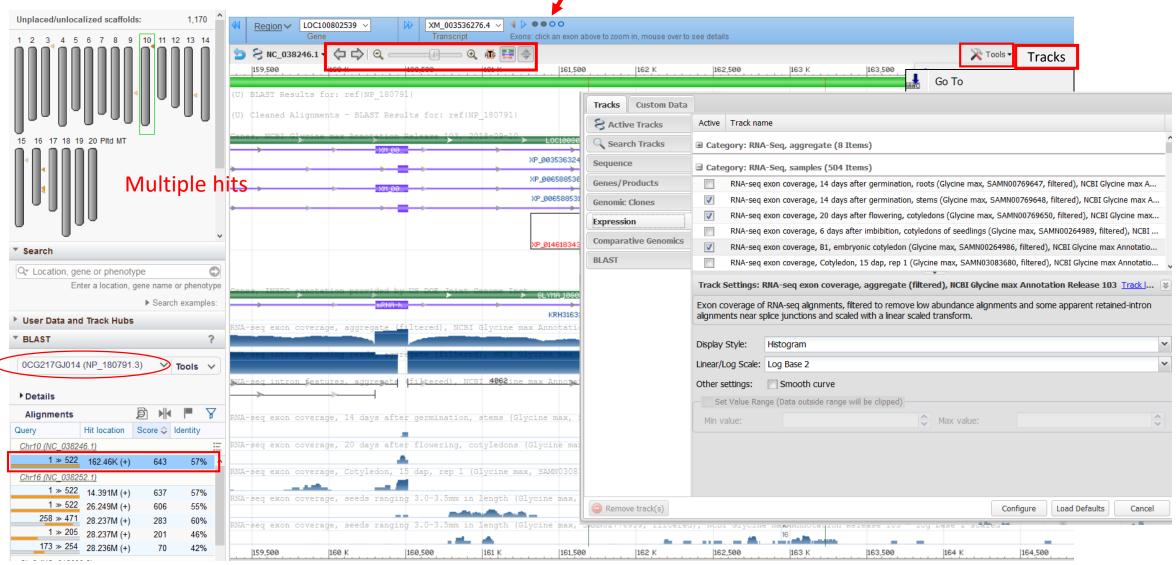


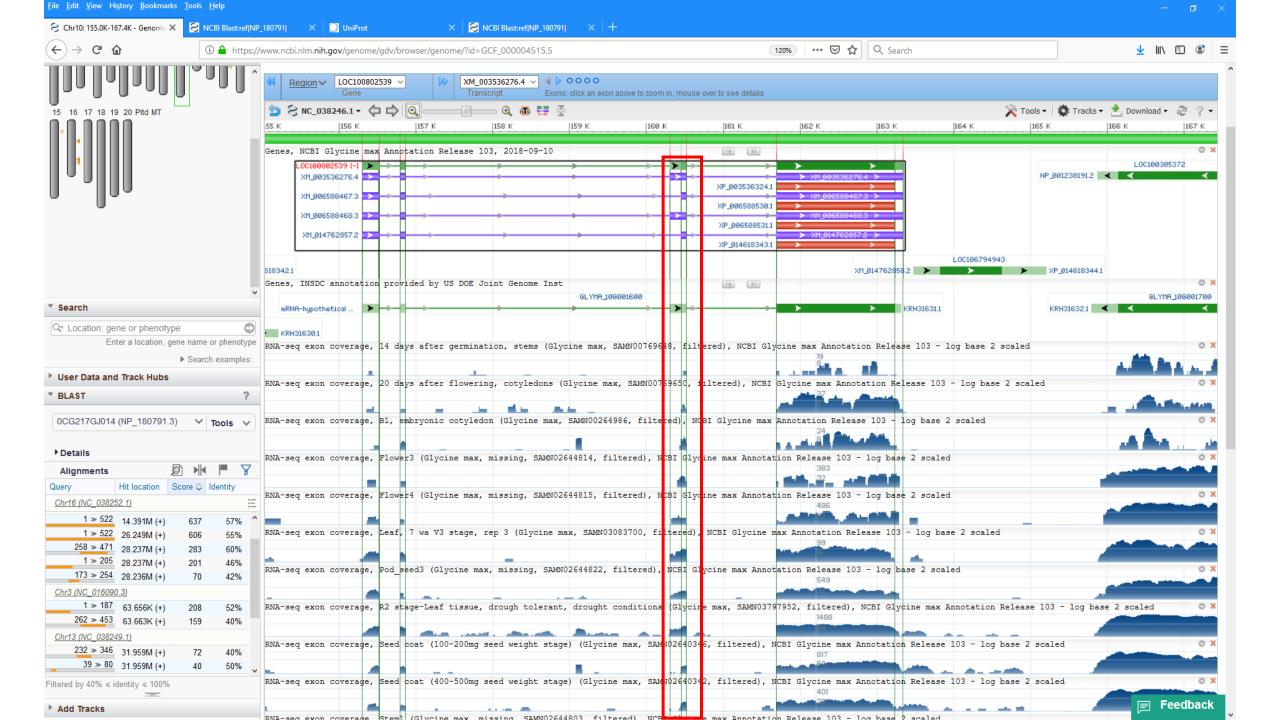


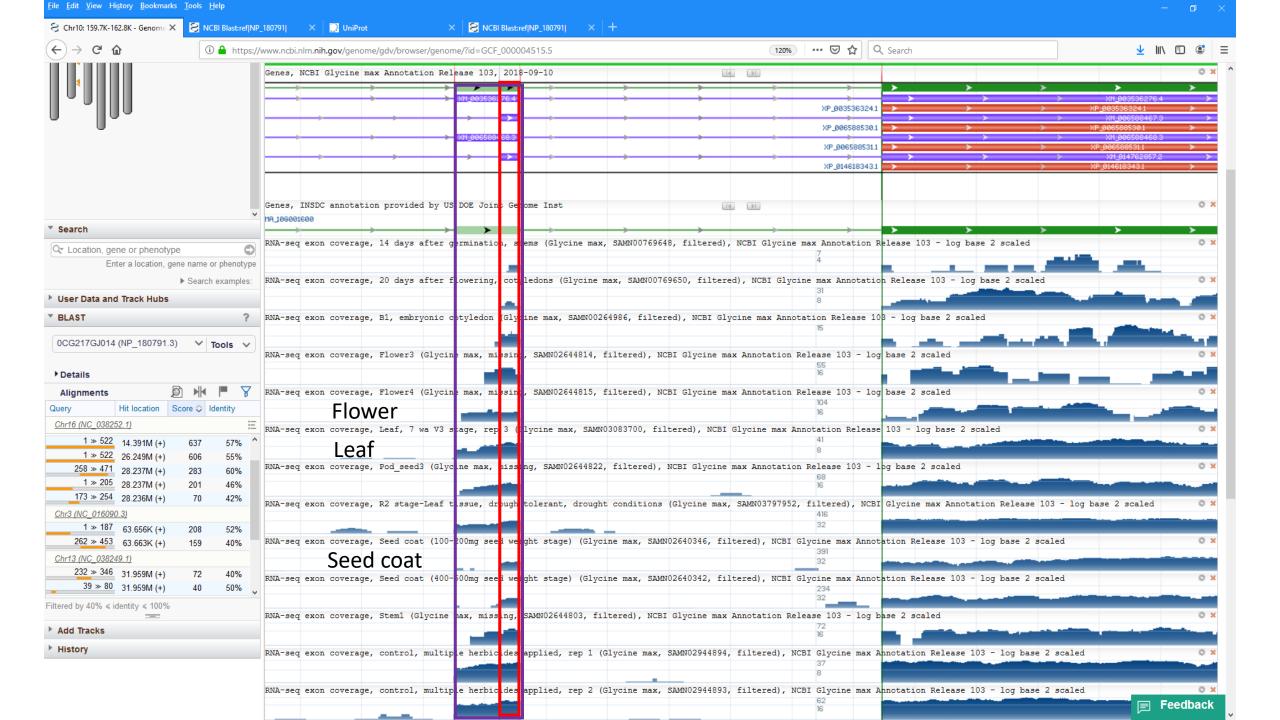












Data Quality

GeneID: 100801486 YIF1B-like protein



*Li et. al., while assessing flavonoid biosynthesis pathway (FBP) genes in various Solanaceae species found that NCBI annotations were generally longer and concluded that

"Overall, the large majority of FBP homologs detected from these various annotation sources were in close agreement, but when they differed, homologs from the NCBI annotations were generally longer and more abundant than from the genome-specific databases. These results suggest that reannotation of genome sequences using a unified annotation strategy, such as employed by the NCBI pipeline, may be preferable for improved consistency during comparative genomics research."

<u>*Li et. al. Genes (Basel).</u> 2019 Jul 25;10(8). Assessing Anthocyanin Biosynthesis in Solanaceae as a Model Pathway for Secondary Metabolism.

Thank you.

RefSeq/Gene

Terence Murphy

Eric Cox

Catherine Farrell

Tamara Goldfarb

Diana Haddad

John Jackson

Vinita Joardar

Kelly McGarvey

Michael Murphy

RefSeq Developers

Alex Astashyn

Olga Ermolaeva

Vamsi Kodali

Craig Wallin

Annotation Pipeline

Francoise Thibaud-Nissen

Paul Kitts Mike Dicuccio

Avi Kimchi

Jinna Choi

Boris Kiryutin

Eyal Mozes

Dan Rausch

Robert Smith

Anton Perkov

Patrick Masterson

Lillian Riddick Wratko Hlavina

Barbara Robbertse Brian Smith-White

Sanjida Rangwala

Pooja Strope

Shashi Pujar

Bhanu Rajput

David Webb

GDV/Remap/GBench

Valerie Schneider

Peter Meric Nathan Bouk

Hsiu-Chuan Chen Cliff Clausen

Anatoliy Kuznetsov

A cast of thousands

Ken Katz

Michael Ovetsky

Lukas Wagner Andrei Shkeda

Donna Maglott

Kim Pruitt

Jim Ostell

Alexandre Souvorov

Watch NCBI News for updates!

http://www.ncbi.nlm.nih.gov/news/

https://www.youtube.com/user/NCBINLM



This research was supported by the Intramural Research Program of the NIH, National Library of Medicine.

NCBI Genome Resources Workshop

Monday January 13, 2020, 12:50 – 3:00 pm, Pacific Salon 1

Time	Topic
12:55 – 1:15	NCBI Wants Your Sequence Data! How Do I Get It There? Ilene Mizrachi
1:15 – 1:35	Annotation of Eukaryote Genomes at NCBI Jinna Hoffman
1:35 – 1:55	Accessing Homologous Gene Datasets at NCBI Nuala O'Leary
1:55 – 2:15	The New PubMed Is Here! Kathi Canese
2:15 – 2:35	Taxonomy Lookup; Data Retrieval: How to Find and Stream Genomic Data in the Cloud! Ben Busby

Visit NCBI Booth 321

Contact us info@ncbi.nlm.nih.gov









Also from NCBI!

Visit NCBI Booth 321 Contact us info@ncbi.nlm.nih.gov

Day	Time	Topic
Saturday	PENDING Royal Palm Salon 3-4	PENDING Aquaculture
Sunday	10:30 am Town & Country	Stand-Alone PGAP: The NCBI Open-Source Pipeline for the Annotation of Prokaryotic Genomes Computational Gene Discovery
Sunday	12:25 pm San Diego	Genomic Resources for Agricultural Animals at NCBI Cattle/Sheep/Goat 2
Sunday	1:42 pm Pacific Salon 1	NCBI RefSeq Resources for Plant Genomics Functional Genomics
Monday	12:50 pm – 3:00 pm <i>Pacific Salon 1</i>	NCBI Genome Resources Workshop
Tuesday	11:10 am California	NCBI BLAST: Enhanced Web Usability through New Result Page and Effective Genomic Data Access Digital Tools and Resources Session 3
Wednesday	11:50 am <i>California</i>	Federated Cloud Access to Datasets through Indexing and/or Graphs! Digital Tools and Resources Session 4



